

What is claimed is:

- 1           1.       A polynucleotide that is regulated by a polypeptide comprising:  
2           a regulatable, catalytically active polynucleotide, wherein the peptide interacts with  
3           the polynucleotide to affect its catalytic activity.
- 1           2.       The polynucleotide of claim 1, wherein the polypeptide is further defined as  
2           being a protein.
- 1           3.       The polynucleotide of claim 1, wherein the polypeptide comprises a peptide of  
2           between about 7 and 20 amino acids.
- 1           4.       The polynucleotide of claim 1, wherein the polypeptide comprises a peptide of  
2           between about 7 and 12 amino acids.
- 1           5.       The polynucleotide of claim 1, wherein the catalytic activity of the nucleic  
2           acid is specific for a nucleic acid target sequence.
- 1           6.       The polynucleotide of claim 1, wherein the catalytic activity of the nucleic  
2           acid is regulated by the interaction of the nucleic acid with an effector.
- 1           7.       The polynucleotide of claim 1, wherein the polynucleotide comprises RNA.
- 1           8.       The polynucleotide of claim 1, wherein the polynucleotide comprises DNA
- 1           9.       The polynucleotide of claim 1, wherein the polynucleotide is at least partially  
2           single stranded.
- 1           10.      The polynucleotide of claim 1, wherein the polynucleotide is at least partially  
2           double stranded.
- 1           11.      The polynucleotide of claim 1, wherein the polynucleotide comprises at least  
2           one modified base.
- 1           12.      The polynucleotide of claim 1, wherein the peptide is endogenous.
- 1           13.      The polynucleotide of claim 1, wherein the peptide is exogenous.
- 1           14.      The polynucleotide of claim 1, wherein the peptide comprises a  
2           phosphorylated peptide.
- 1           15.      A nucleic acid that is regulated by an effector comprising:  
2           a regulatable, catalytically active nucleic acid, generated by the modification of at  
3           least one catalytic residue.

1           16.     The nucleic acid of claim 15, wherein the catalytic activity of the nucleic acid  
2 is specific for a nucleic acid target sequence.

1           17.     The nucleic acid of claim 15, wherein the catalytic activity of the nucleic acid  
2 is regulated by the interaction of the nucleic acid with an effector.

1           18.     The nucleic acid of claim 15, wherein the nucleic acid comprises RNA.

1           19.     The nucleic acid of claim 15, wherein the nucleic acid comprises DNA.

1           20.     The nucleic acid of claim 15, wherein the nucleic acid is at least partially  
2 single stranded.

1           21.     The nucleic acid of claim 15, wherein the nucleic acid is at least partially  
2 double stranded.

1           22.     The nucleic acid of claim 15, wherein the nucleic acid comprises at least one  
2 modified base.

1           23.     The nucleic acid of claim 15, wherein the effector is endogenous.

1           24.     The nucleic acid of claim 15, wherein the effector is exogenous.

1           25.     The nucleic acid of claim 15, wherein the effector comprises a protein.

1           26.     The nucleic acid of claim 15, wherein the effector comprises a pharmaceutical  
2 agent.

1           27.     The nucleic acid of claim 15, wherein the effector comprises a protein  
2 complex.

1           28.     The nucleic acid of claim 15, wherein the effector comprises a peptide.

1           29.     The nucleic acid of claim 15, wherein the effector a phosphorylated peptide.

1           30.     The nucleic acid of claim 15, wherein the effector comprises a  
2 dephosphorylated peptide.

1           31.     The nucleic acid of claim 15, wherein the nucleic acid catalyses a reaction that  
2 causes the expression of a target gene to be up-regulated.

1           32.     The nucleic acid of claim 15, wherein the nucleic acid catalyses a reaction that  
2 causes the expression of a target gene to be down-regulated.

1           33.     The nucleic acid of claim 15, wherein the nucleic acid is used to detect at least  
2 one exogenous effector from a library of candidate exogenous effector molecules.

1           34.     The nucleic acid of claim 15, wherein the nucleic acid and the effector form a  
2 nucleic acid-effector complex.

1           35.     The nucleic acid of claim 15, wherein the nucleic acid and the effector is a  
2 molecule that forms an nucleic acid-effector complex and the nucleic acid-effector complex  
3 acts synergistically to affect the catalytic activity of the nucleic acid-effector complex.

1           36.     The nucleic acid of claim 15, wherein the nucleic acid catalyses a ligation  
2 reaction with an oligonucleotide substrate.

1           37.     The nucleic acid of claim 15, wherein the nucleic acid catalyses a reaction that  
2 adds a non-oligonucleotide substrate.

1           38.     The nucleic acid of claim 15, wherein the nucleic acid catalyses a reaction that  
2 adds biotin to the nucleic acid.

1           39.     The nucleic acid of claim 15, wherein the nucleic acid catalyses a cleavage  
2 reaction with an oligonucleotide substrate.

1           40.     The nucleic acid of claim 15, in which the kinetic parameters of nucleic acid  
2 catalysis are altered in the presence of one or more effector-effectors that acts on the effector  
3 molecule that interacts with the nucleic acid.

1           41.     The nucleic acid of claim 15, in which the kinetic parameters of nucleic acid  
2 catalysis are altered in the presence of theophylline.

1           42.     The nucleic acid of claim 15, in which the kinetic parameters of nucleic acid  
2 catalysis are altered in the presence of a supermolecular structure.

1           43.     The nucleic acid of claim 15, in which the kinetic parameters of nucleic acid  
2 catalysis are altered in the presence of a supermolecular structure that comprises a virus  
3 particle.

1           44.     The nucleic acid of claim 15, in which the kinetic parameters of nucleic acid  
2 catalysis are altered in the presence of a supermolecular structure that comprises a cell wall.

1           45.     A nucleic acid comprising:

2 a gene;

3 a regulatable, catalytically active nucleic acid inserted within the gene;

4 wherein the presence of an effector causes the nucleic acid to catalyze a reaction.

1           46.     The nucleic acid of claim 45, wherein the catalytic reaction is a self-splicing  
2 reaction.

1           47.     The nucleic acid of claim 45, wherein the catalytic reaction is a ligation  
2 reaction.

1           48.     The nucleic acid of claim 45, wherein the catalytic reaction is a trans-cleavage  
2 reaction.

1           49.     The nucleic acid of claim 45, wherein the catalytic activation of the nucleic  
2 acid leads to changes in expression of the gene.

1           50.     The nucleic acid of claim 45, wherein the catalytic activation of the nucleic  
2 acid leads to changes in expression of one or more genes.

1           51.     The nucleic acid of claim 45, wherein the catalytic activation of the nucleic  
2 acid leads to changes in expression of the mRNA of the gene.

1           52.     The nucleic acid of claim 45, wherein the catalytic activation of the nucleic  
2 acid leads to changes in expression of the protein encoded by the gene.

1           53.     A nucleic acid segment comprising:  
2           a regulatable, catalytically active nucleic acid comprising one or more catalytic  
3 nucleotides, selected from a pool of nucleic acids in which at least one of the catalytic  
4 residues has been randomized.

1           54.     A regulatable, catalytically active nucleic acid segment comprising:  
2           an effector domain; and  
3           a nucleic acid catalyst domain in which one or more catalytic residues of the nucleic  
4 acid catalyst have been randomized;  
5           wherein the kinetic parameters of the catalytic domain are regulated by an effector  
6 that interacts with the effector domain.

1           55.     A method of isolating a regulatable, catalytically active nucleic acid,  
2 comprising the steps of:  
3           randomizing at least one nucleotide in the catalytic domain of a catalytically active  
4 nucleic acid to create a nucleic acid pool; and  
5           removing from the nucleic acid pool those nucleic acids that interact with the  
6 catalytic target of the catalytic domain.

1           56.     The method of claim 55, further comprising the step of adding an effector to  
2 the remaining pool of nucleic acids.

1           57.     The method of claim 55, further comprising the steps of adding an effector to  
2 the remaining nucleic acids, wherein the effector acts on the nucleic acids to alter the  
3 catalytic activities of the nucleic acids.

1           58.     The method of claim 55, further comprising the step of purifying the isolated  
2     nucleic acid.

1           59.     The method of claim 55, further comprising the step of sequencing the  
2     isolated nucleic acid.

1           60.     The method of claim 55, wherein the step of removing the nucleic acids is  
2     under high stringency conditions.

1           61.     The method of claim 55, wherein the step of removing the nucleic acids is  
2     under moderate stringency conditions.

1           62.     The method of claim 55, wherein the step of removing the nucleic acids is  
2     under low stringency conditions.

1           63.     The method of claim 55, where the target is an mRNA molecule.

1           64.     The method of claim 56, where the effector is a protein.

1           65.     The method of claim 56, where the effector is a peptide.

1           66.     The method of claim 56, where the effector is a phosphoprotein.

1           67.     The method of claim 56, where the effector is a glycoprotein.

1           68.     The method of claim 56, where the effector is light.

1           69.     The method of claim 56, where the effector is visible light.

1           70.     The method of claim 56, where the effector is a magnet.

1           71.     The method of claim 55, where the target is a metabolic reaction.

1           72.     The method of claim 55, in which nucleic acids with altered catalytic  
2     specificity are selected in the presence of an effector.

1           73.     The method of claim 55, in which nucleic acids with altered catalytic  
2     activities are selected in the absence of an effector.

1           74.     The method of claim 55, in which nucleic acids with altered catalytic  
2     activities are serially selected in the presence and the absence of an effector.

1           75.     The method of claim 55, the effector domain comprises a random sequence  
2     pool.

1           76.     The method of claim 55, the effector domain comprises a partially randomized  
2     sequence pool.

1           77.     A method of making a regulatable, catalytically active nucleic acid,  
2     comprising the steps of:

3 contacting a pool of nucleic acids, the nucleic acids having a catalytic and an effector  
4 domain, wherein at least one nucleotide in the catalytic domain of the nucleic acids has been  
5 randomized;

6 removing from the nucleic acid pool those nucleic acids that interact with the  
7 catalytic target of the catalytic domain;

8 adding an effector protein to the remaining nucleic acids; and

9 isolating those nucleic acids that interact with the catalytic target of the catalytic  
10 domain.

1 78. A method of isolating a regulatable, catalytically active nucleic acid,  
2 comprising the steps of:

3 randomizing at least one nucleotide in the catalytic domain of a catalytically active  
4 nucleic acid to create a nucleic acid pool;

5 removing from the nucleic acid pool those nucleic acids that interact with the  
6 catalytic target of the catalytic domain;

7 adding an effector molecule to the nucleic acids; and

8 isolating those nucleic acids that interact with the catalytic target of the catalytic  
9 domain.

1 79. A method of isolating a regulatable, catalytically active nucleic acid having a  
2 catalytic and an effector domain, comprising the steps of:

3 randomizing at least one nucleotide in the catalytic domain of the nucleic acid to  
4 create a nucleic acid pool;

5 removing from the nucleic acid pool those randomized nucleic acids that interact with  
6 the catalytic target of the catalytic domain;

7 adding an effector to the nucleic acids; and

8 isolating the nucleic acids that interact with the catalytic target of the catalytic  
9 domain.

1 80. An automated method of isolating a regulatable, catalytically active nucleic  
2 acid having a catalytic and an effector domain, comprising the steps of:

3 (a) randomizing at least one nucleotide in the catalytic domain of the nucleic acid  
4 to create a nucleic acid pool;

- 5 (b) removing from the nucleic acid pool those randomized nucleic acids that  
6 interact with the catalytic target of the catalytic domain;  
7 (c) adding an effector to the nucleic acids;  
8 (d) adding an effector-effector that specifically interacts with the effector; and  
9 (e) isolating the nucleic acids that interact with the catalytic target of the catalytic  
10 domain; and  
11 (f) repeating steps (a) through (e).

1 81. A method of detection of a target using a regulatable, catalytically active  
2 nucleic acid comprising the steps of:  
3 contacting the a regulatable, catalytically active nucleic acid with the target; and  
4 measuring the effect of the interaction between the a regulatable, catalytically active  
5 nucleic acid and the target.

1 82. A method of modifying a target using a regulatable, catalytically active  
2 nucleic acid comprising the steps of:  
3 providing a regulatable, catalytically active nucleic acid capable of target specific  
4 modification; and  
5 modifying the target under conditions that cause a regulatable, catalytically active  
6 nucleic acid-specific activity.

1 83. A biosensor comprising:  
2 a solid support; and  
3 at least one regulatable, catalytically active nucleic acid, wherein the kinetic  
4 parameters of the nucleic acid on a target vary in response to the interaction of an effector  
5 molecule with the nucleic acid;  
6 wherein the at least one regulatable, catalytically active nucleic acid is immobilized  
7 on the support.

1 84. The biosensor of claim 83, wherein the reaction is machine readable.

1 85. The biosensor of claim 83, wherein the solid support comprises a multiwell  
2 plate.

1 86. The biosensor of claim 83, wherein the solid support comprises a surface  
2 plasmon resonance sensor.

1           87.     The biosensor of claim 83, wherein the at least one regulatable, catalytically  
2 active nucleic acids is covalently immobilized on the solid support.

1           88.     The biosensor of claim 83, wherein the catalytic reaction produces a  
2 detectable signal.

1           89.     The biosensor of claim 83, wherein the catalytic reaction is the attachment of  
2 a tag to the immobilized nucleic acids to produce the signal.

1           90.     The biosensor of claim 83, wherein the substrate is further defined as  
2 containing known nucleic acid sequences tags and the nucleic acids are sorted on the surface  
3 of the substrate based on non-covalent hybridization to sequence tags.

1           91.     A biosensor comprising:  
2 a solid support; and  
3 at least one regulatable, catalytically active nucleic acids, wherein the kinetic  
4 parameters of the nucleic acids on a target vary in response to the interaction of an effector  
5 molecule with the nucleic acid;

6 wherein catalytic targets of the catalytic domain is immobilized on the support.

1           92.     A biosensor comprising:  
2 a solid support; and  
3 at least one regulatable, catalytically active nucleic acids, wherein the kinetic  
4 parameters of the nucleic acids on a target vary in response to the interaction of an effector  
5 molecule with the nucleic acid;

6 wherein the effector is immobilized on the support.

1           93.     A method of selecting a regulatable, catalytically active nucleic acid,  
2 comprising the steps of:  
3 contacting a pool of nucleic acids, the nucleic acids having a catalytic and an effector  
4 domain, wherein at least one nucleotide in the catalytic domain of the nucleic acids has been  
5 randomized;

6 removing from the nucleic acid pool those nucleic acids that interact with the  
7 catalytic target of the catalytic domain;

8 adding an effector to the remaining nucleic acids; and

9 isolating those nucleic acids that interact with the catalytic target of the catalytic  
10 domain;



11 introducing the nucleic acids into a host cell; and  
12 measuring the catalytic activity of the nucleic acid upon exposure of the host cell to  
13 the effector.

1 94. The method of claim 93, further comprising the step of purifying the isolated  
2 nucleic acid.

1 95. The method of claim 93, further comprising the step of sequencing the  
2 isolated nucleic acid.

1 96. The method of claim 93, wherein the step of removing the nucleic acids is  
2 under high stringency conditions.

1 97. The method of claim 93, wherein the step of removing the nucleic acids is  
2 under moderate stringency conditions.

1 98. The method of claim 93, wherein the step of removing the nucleic acids is  
2 under low stringency conditions.

1 99. The method of claim 93, where the target is an mRNA molecule.

1 100. The method of claim 93, where the effector is a protein.

1 101. The method of claim 93, where the effector is a peptide.

1 102. The method of claim 93, where the effector is a phosphoprotein.

1 103. The method of claim 93, where the effector is a glycoprotein.

1 104. The method of claim 93, where the effector is light.

1 105. The method of claim 93, where the effector is visible light.

1 106. The method of claim 93, where the effector is a magnet.

1 107. The method of claim 93, in which nucleic acids with altered catalytic  
2 activities are serially selected in the presence and the absence of the effector.

1 108. The method of claim 93, the effector domain comprises a completely random  
2 sequence pool.

1 109. The method of claim 93, the effector domain comprises a partially randomized  
2 sequence pool.

1 110. A method of selecting a regulatable, catalytically active nucleic acid,  
2 comprising the steps of:

3 contacting a pool of nucleic acids, the nucleic acids having a catalytic and an effector  
4 domain, wherein at least one nucleotide in the catalytic domain of the nucleic acids has been  
5 randomized;

6 removing from the nucleic acid pool those nucleic acids that interact with the  
7 catalytic target of the catalytic domain;

8 adding an effector to the remaining nucleic acids; and

9 isolating those nucleic acids that interact with the catalytic target of the catalytic  
10 domain;

11 introducing the nucleic acids into a host cell; and

12 measuring the catalytic activity of the nucleic acid upon exposure of the host cell to  
13 the effector.

1 111. The method of claim 110, further comprising the step of purifying the isolated  
2 nucleic acid.

1 112. The method of claim 110, further comprising the step of sequencing the  
2 isolated nucleic acid.

1 113. The method of claim 110, wherein the step of removing the nucleic acids is  
2 under high stringency conditions.

1 114. The method of claim 110, wherein the step of removing the nucleic acids is  
2 under moderate stringency conditions.

1 115. The method of claim 110, wherein the step of removing the nucleic acids is  
2 under low stringency conditions.

1 116. The method of claim 110, where the target is an mRNA molecule.

1 117. The method of claim 110, where the effector is a protein.

1 118. The method of claim 110, where the effector is a peptide.

1 119. The method of claim 110, where the effector is a phosphoprotein.

1 120. The method of claim 110, where the effector is a glycoprotein.

1 121. The method of claim 110, where the effector is light.

1 122. The method of claim 110, where the effector is visible light.

1 123. The method of claim 110, where the effector is a magnet.

1 124. The method of claim 110, in which nucleic acids with altered catalytic  
2 activities are serially selected in the presence and the absence of the effector.

1           125. The method of claim 110, the effector domain comprises a completely random  
2 sequence pool.

1           126. The method of claim 110, the effector domain comprises a partially  
2 randomized nucleotide sequence.

1           127. A method of detecting a regulatable, catalytically active nucleic acid,  
2 comprising the steps of:

3           isolating a regulatable, catalytically active nucleic acid;

4           creating a construct in which the nucleic acid is in position to regulate the expression  
5 of a reporter gene;

6           introducing the construct into a host cell; and

7           measuring the catalytic activity of the nucleic acid upon exposure of the host cell to  
8 an effector.

1           128. A vector comprising:

2           a regulatable, catalytically active polynucleotide, wherein the peptide molecule  
3 interacts with the polynucleotide to affect its catalytic activity.

1           129. A vector comprising:

2           a regulatable, catalytically active nucleic acid, generated by the modification of at  
3 least one catalytic residue.

1           130. A method of modulating expression of a nucleic acid, the method comprising

2           providing a polynucleotide that is regulated by a peptide, the polynucleotide

3           comprising a regulatable, catalytically active polynucleotide, wherein the peptide interacts  
4 with the polynucleotide to affect its catalytic activity; and

5           contacting the polynucleotide with the peptide, thereby modulating expression of a  
6 nucleic acid.

1           131. The method of claim 130, wherein the polynucleotide is provided in a cell.

1           132. The method of claim 131, wherein the cell is provided in vitro.

1           133. The method of claim 131, wherein the cell is provided in vivo.

1           134. The method of claim 131, wherein the cell is a prokaryotic cell.

1           135. The method of claim 131, wherein the cell is a eukaryotic cell.

1           136. A method of modulating expression of a nucleic acid, the method comprising  
2 the steps of:

- 3 providing a nucleic acid that is regulated by an effector, the nucleic acid comprising:  
4 a regulatable, catalytically active nucleic acid, wherein the regulatable, catalytically active  
5 nucleic acid molecule includes at least one modified catalytic residue; and  
6 contacting the nucleic acid with the effector, thereby modulating expression of a  
7 nucleic acid.